

Weightlessness and The Human Skeleton: A New Perspective

Michael F. Holick, Ph.D., M.D.
 Director, General Clinical Research Center
 Boston University School of Medicine
 Boston, Massachusetts

57-52

207616

P-16

Introduction

It is now clear after more than two decades of space exploration that one of the major short- and long-term effects of microgravity on the human body is the loss of bone. The purpose of this presentation will be to review the data regarding the impact of microgravity and bed rest on calcium and bone metabolism. I will take the position in this Socratic debate that the effect of microgravity on bone metabolism can either be reversed or be mitigated.

As we begin to contemplate long-duration space flight, habitation of Space Station Freedom and the Moon, one of the issues that will need to be addressed is whether humans need to maintain a skeleton that has been adapted for the one-g force on Earth? Clearly in the foreseeable future a healthy and structurally sound skeleton will be required for astronauts to shuttle back and forth from Earth to the Moon, Space Station, and Mars. Based on most available data from bed-rest studies and the short- and long-duration microgravity experiences by astronauts and cosmonauts, bone loss is a fact of life in this environment. With the rapid advances in our understanding of bone physiology is it now possible to contemplate measures that can prevent or mitigate microgravity-induced bone loss. Will the new therapeutic approaches for enhancing bone mineralization be useful for preventing significant bone loss during long-term space flight? Are there other approaches such as exercise and electrical stimulation that can be used to mitigate the impact of microgravity on the skeleton? A recent study that evaluated the effect of microgravity on bone modeling in developing chick embryos may perhaps provide a new perspective about the impact of microgravity on bone metabolism.

Human Bone Disease: Historical Perspective and Concerns for the Future

Bone disease has afflicted humans almost since the beginning of time. As the industrial revolution took hold in Northern Europe, rickets became an epidemic. This devastating bone disease was eventually eradicated as significant health problem because of the appreciation of the beneficial effect of sunlight in producing vitamin D in the skin [6]. During the past two decades osteoporosis has become a significant health issue affecting more than 20 million elderly Americans and costing \$6 billion dollars that by the year 2020 we will begin the colonization of the Moon and travel to Mars. This exciting new era in space exploration could result in a new bone disease caused by microgravity. If we are unable to prevent or reverse microgravity-induced bone loss, this negative effect on the skeleton could severely limit human aspirations to explore and colonize the universe.

As we initiate a program for long-term human habitation in a microgravity environment on board Space Station Freedom and the Moon there remains concern about the acute and chronic loss of bone mineral. Acutely the rapid loss of bone mineral in combination with microgravity-induced hemodynamic changes could significantly increase the risk of kidney stones and soft tissue calcification. Chronic unrelenting bone loss could compromise its structural integrity to such a degree that when astronauts are exposed to the G-forces upon reentry into Earth's gravitational field, micro- and macrofractures of the skeletal network could occur. In addition, since the entire skeleton is unloaded in microgravity, it is possible that its three-dimensional structure is

reorganized to such a degree that when astronauts return to Earth and initiate routine activity in a one-g environment that micro- and macrofractures of the skeleton could occur as the body realigns the framework of the skeleton to adapt to the forces put upon it by Earth's gravity.

The Effect of Bed Rest and Microgravity on Bone Metabolism

The model that is most often used to evaluate the potential impact of microgravity on bone and calcium metabolism is to unload the skeleton of healthy subjects by putting them at strict bed rest [4, 9, 14, 20]. Schneider and McDonald [14] and Arnaud, Schneider, and Morey-Holton [1] measured intestinal calcium transport 4 weeks before and 20 weeks during strict bed rest. They showed the efficiency of intestinal calcium absorption gradually declined during the first 4 weeks of bed rest and plateaued to about 50 percent of the original level during that last 16 weeks (Figure 1).

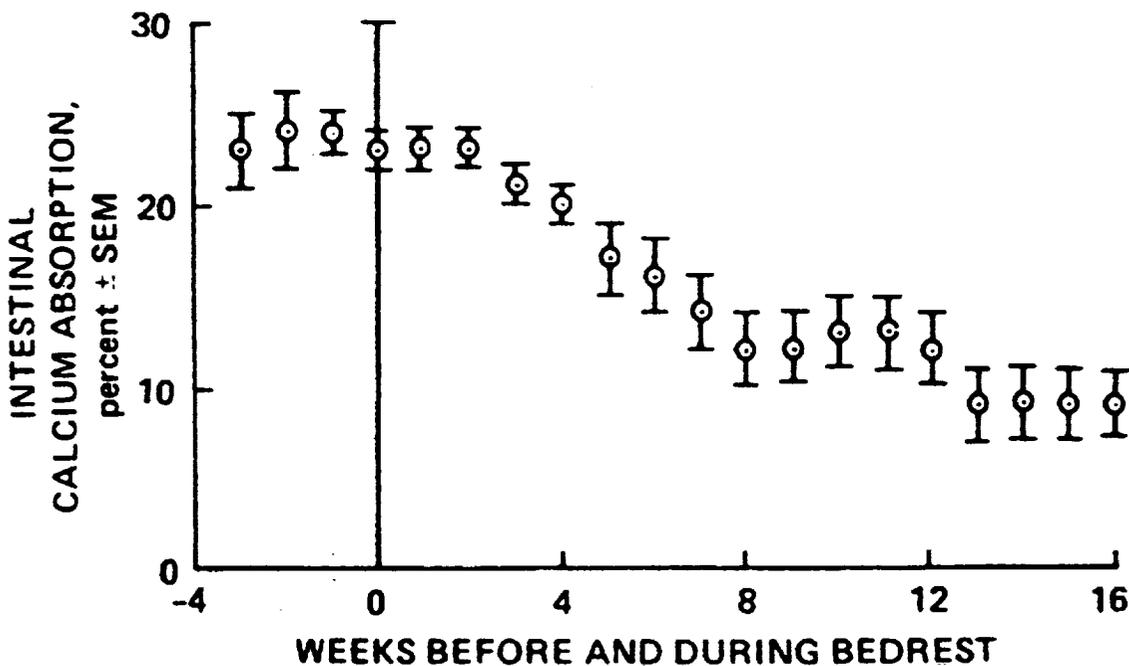


Figure 1. *The intestinal absorption of calcium estimated from balance data in 19 to 47 healthy subjects immobilized by bed rest for 16 weeks. Absorption was determined in subjects consuming 1000 mg daily (reproduced with permission).*

An increase in the mobilization of calcium from the skeleton may transiently increase the circulating ionized calcium concentrations which in turn caused a decrease in parathyroid hormone (PTH) secretion [1, 16]. A decrease in PTH secretion would result in the renal wasting of calcium causing an increased urinary output of calcium (Figure 2). The decrease in intestinal calcium absorption, that was most likely caused by a decrease in the PTH mediated metabolism of 25-hydroxyvitamin D to 1,25-dihydroxyvitamin D, resulted in an increase loss of calcium in the feces (Figure 2). This caused a negative calcium balance of approximately 200 mg/day during the last 16 weeks of bed rest (Figure 2).

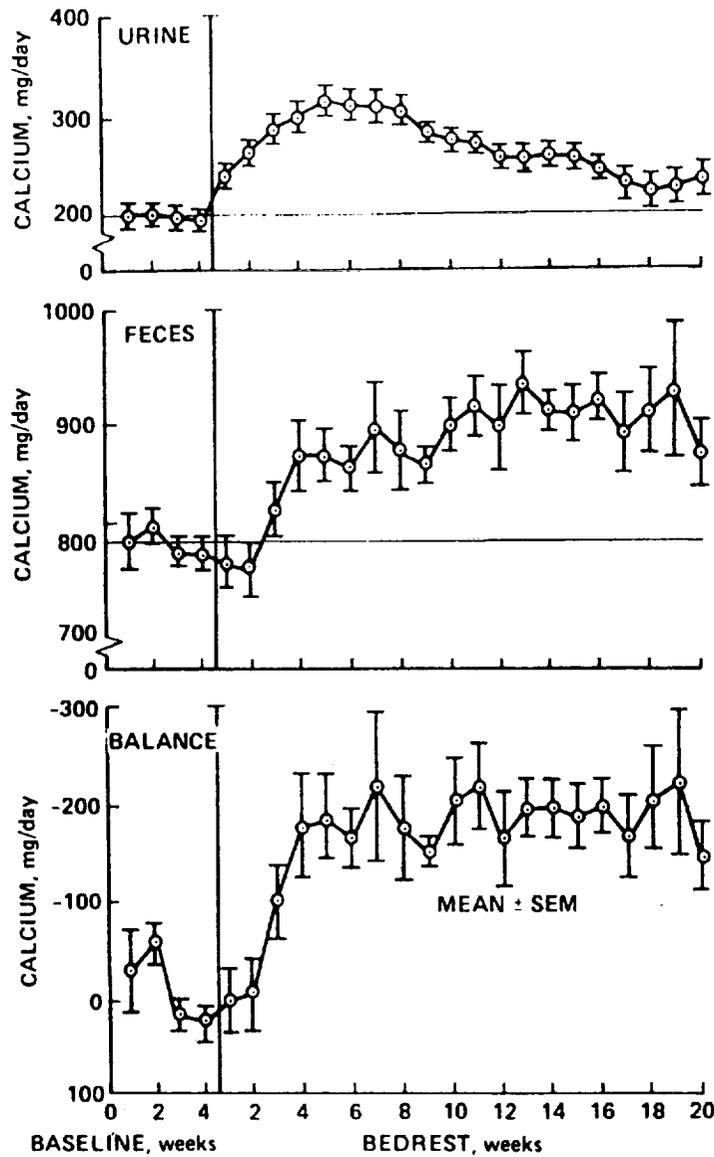


Figure 2. *The excretion of calcium in the urine and feces, in milligrams per day, in 12 to 32 healthy subjects immobilized by bed rest for 20 weeks. Dietary calcium was kept constant in these subjects at 1000 mg/d. Fecal calcium was corrected for time by polyethylene glycol 4000 administered three times daily in a dose 500 mg. The lowest panel shows the results of the balance study estimated from the intake and excretion of calcium in the urine and feces (reproduced with permission).*

Urinary hydroxyproline concentrations were increased providing evidence that the increase loss of calcium in the urine was partly due to an increase turn-over of the bone (Figure 3).

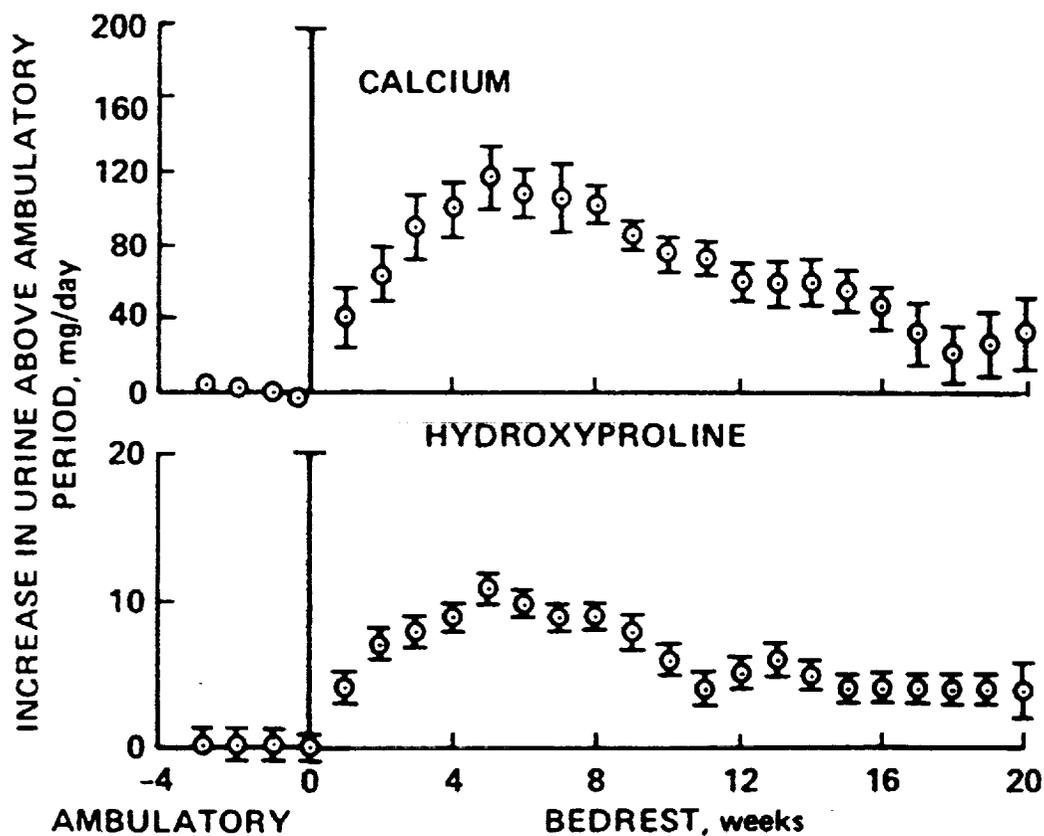


Figure 3. *The daily urinary excretion of hydroxyproline and calcium in the urine. Excretion is shown as the percentage of increase from the ambulatory control period of 4 weeks in 14 to 40 normal subjects during 20 weeks of bed rest (reproduced with permission).*

Bone density measurements of the calcaneus from the bed-rest subjects showed a marked decline in bone mineral density. A significant decrease in the calcaneal bone density was evident by 5 to 8 weeks and continued to decline at about the same rate for the next 20 weeks. Approximately 30 percent of the bone mineral density was lost by the end of the study (Figure 4).

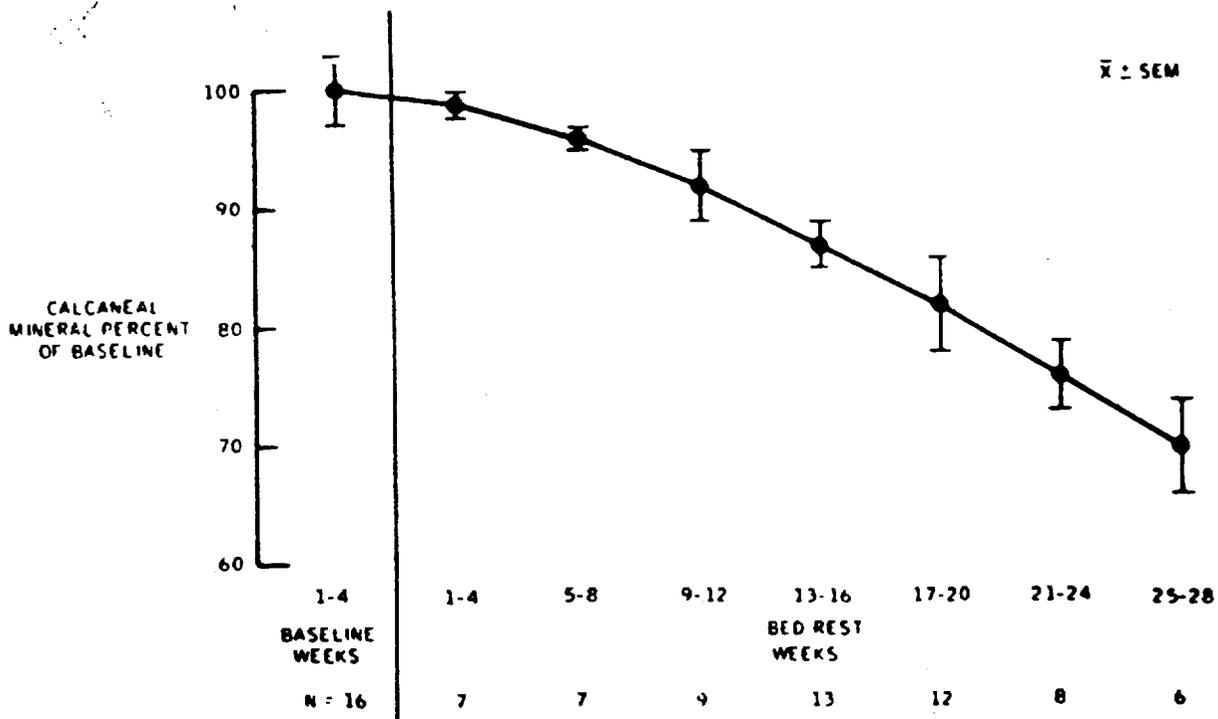


Figure 4. *Photon absorptiometry of the calcaneous. Percent decrease from ambulatory values during untreated bed rest (reproduced with permission).*

The results of these bed-rest studies have mirrored the observations seen in astronauts and cosmonauts exposed to microgravity. Rambaut, Leach, and Whedon [11] observed significant increases in urinary calcium excretion in Skylab 4 astronauts (Figure 5).

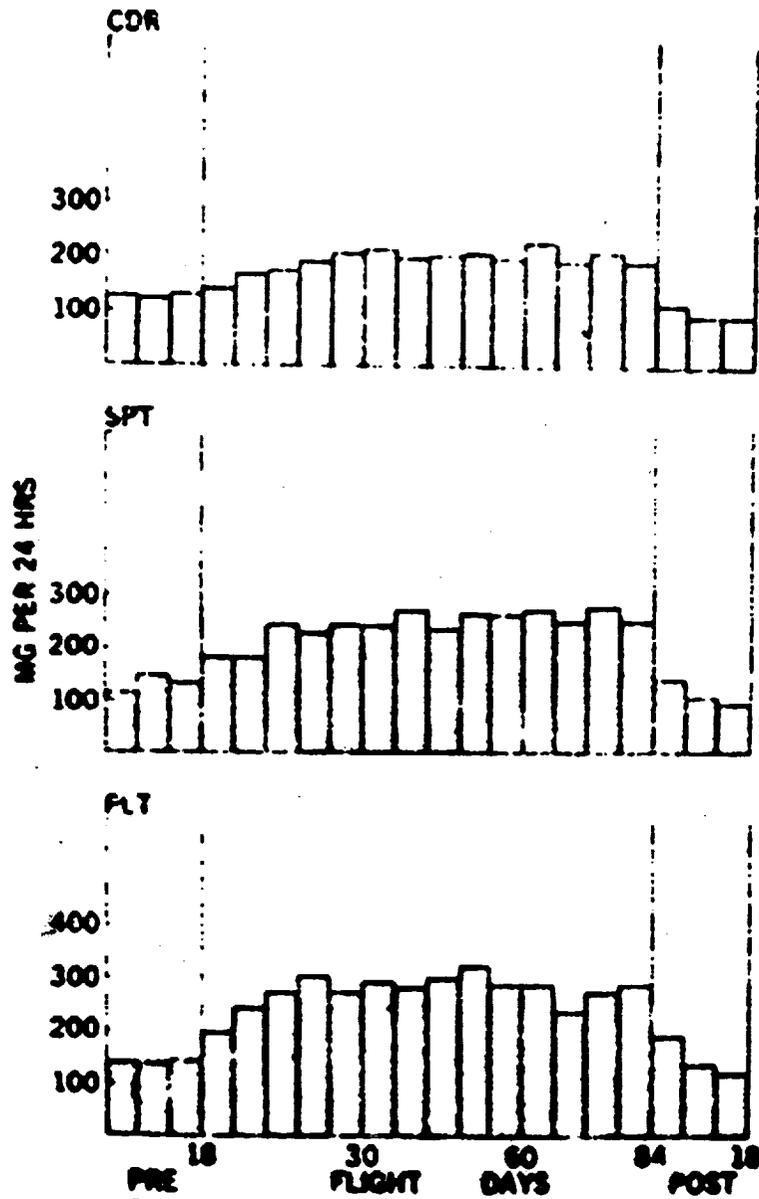


Figure 5. *Urinary calcium in Skylab 4 astronauts (reproduced with permission).*

They also found a significant decrease in calcaneous bone density during the 84-day mission on board Skylab 4 (Figure 6).

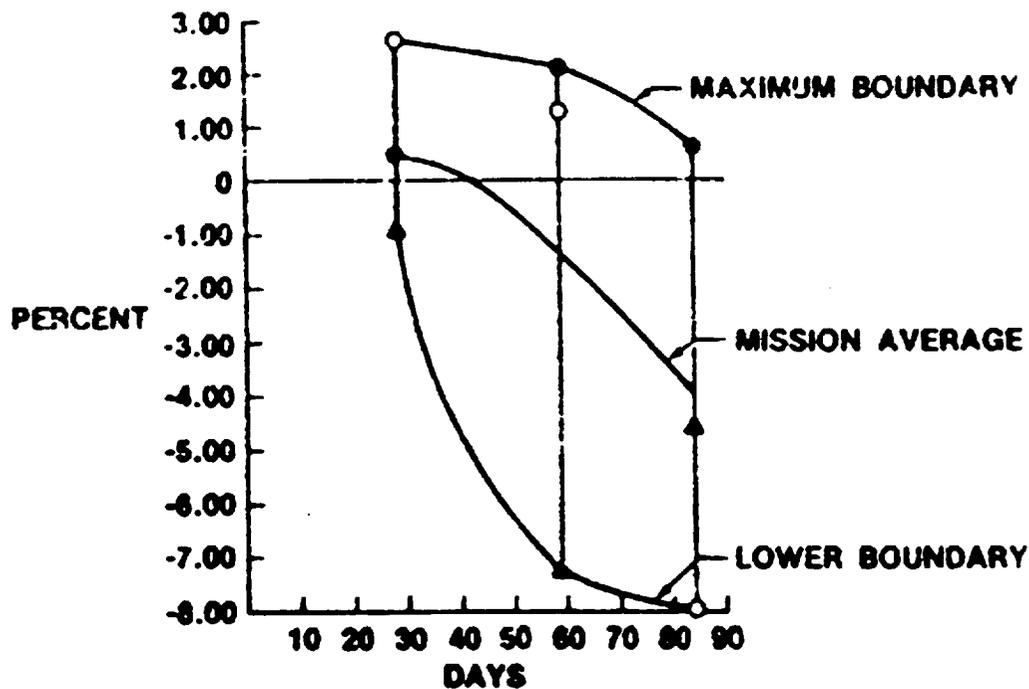


Figure 6. *Calcaneal mineral loss as a function of Skylab flight duration (reproduced with permission).*

Parfitt [10] analyzed the calcaneal mineral bone density in astronauts on board Skylabs 2, 3 and 4 and found a significant decrease of about 4 percent in the longest flights of 59 and 84 days. By contrast no significant changes were noted in the cortical bone density of the radius and ulna in the same astronauts. The Skylab astronauts experienced a negative calcium balance of about 200 to 300 mg/day. Parfitt's analysis of the Skylab 4 astronauts' calcium metabolism data also revealed that they lost about 300 mg of calcium a day that resulted in a total body loss of about 18 g (with a range of 10 to 22 g). Most of the loss was not due to a decrease in intestinal absorption of calcium; it was caused by an increase in urinary excretion of calcium. Parfitt concluded that two-thirds of the total calcium loss came from trabecular bone and one third came from cortical bone. Similar observations have been made in cosmonauts. In Table 1 all of the cosmonauts had significant decrease in the bone density of after 140 days in microgravity [17].

Table 1. *Percentage change in mineral content of calcaneous in cosmonauts*

Crew Member	Flight Duration (days)	Postflight Testing Day	Entire Flight	Monthly Average
CDR	140	5	-3.0	-0.64
FLE	140	4	-19.8	-4.24
CDR	175	5	-9.8	-1.69
FLE	175	5	-3.0	-0.51
CDR	184	14	-9.6	-1.56
FLE	184	14	-4.4	-0.71
CDR	75	4	-0.9	-0.36
FLE	75	4	-3.2	-1.31

Physiologic Function of the Skeleton

In light of the observations just presented there is the possibility that a significant amount of skeletal mass will be lost during long-term space travel. One of the questions that is worthwhile asking is, "Do we need a skeleton?" Is it possible that we will ultimately evolve in microgravity into protoplasmic-thinking beings that do not require a skeleton? Some of the functions of the skeleton are obvious. It protects our soft vital organs, provides attachment for our musculature and permits us to be mobile in our terrestrial environment. A more subtle but equally important reason for the evolution of a calcified skeleton is to provide a storage depot for calcium. As vertebrates evolved in the fertile oceans they were bathed in calcium. When they left this environment for terra firma they were confronted with a calcium deprived environment. They needed to develop mechanisms by which they could efficiently utilize, assimilate, and store the limited supplies of calcium that were in their environment. Vertebrates also developed elaborate mechanisms to call upon these reserves at times of need.

Calcium plays an essential role in neurotransmission and neuromuscular function. In addition, cytosolic calcium concentrations are carefully regulated and are responsible for a multitude of cellular functions. Calcium also acts as an important second messenger to the cell. Therefore, I believe that there is a need to maintain our skeletal structure if for no other reason than to act as a storage site for calcium so that the body can call on this depot to maintain intra- and extracellular calcium concentrations within physiologically acceptable limits.

Dynamics of Bone Formation and Bone Resorption: Strategies for Maintaining Bone Mineral Density in a Microgravity Environment

When bone formation rates are greater than bone degradation rates there is an increase in bone mass whereas, when bone formation rates are less than bone degradation rates there is a decrease in bone mass. Osteoporosis is a state whereby the bone matrix along with the bone mineral is lost. Osteomalacia on the other hand occurs when there is loss of bone mineral with the maintenance of the matrix [8]. Although it is well documented that microgravity causes a decrease in bone mineral content it is not known whether the bone mineral matrix is lost to the same degree causing osteoporosis or whether the matrix is partially or fully maintained causing osteomalacia. If the matrix is maintained in a microgravity environment then there is an opportunity to develop therapeutic strategies to replenish the demineralized matrix. If, on the other hand, microgravity induced bone demineralization causes the loss of the staging network (matrix) and calcium

hydroxyapatite (mineral) that results in the destruction of the bone trabeculae, then it will be very difficult if not impossible to restore the very fine trabecular network. Dempster *et al.* [3] have demonstrated this in an exquisite fashion with scanning electromicroscopy. Figure 7a shows the trabecular pattern from a healthy adult male skeleton while Figure 7b shows a marked loss in the thickness and number of trabeculae as well as gaps between the trabeculae in a woman with osteoporosis and multiple compression fractures of her spine. It would be very difficult to induce bone cells to bridge these gaps.

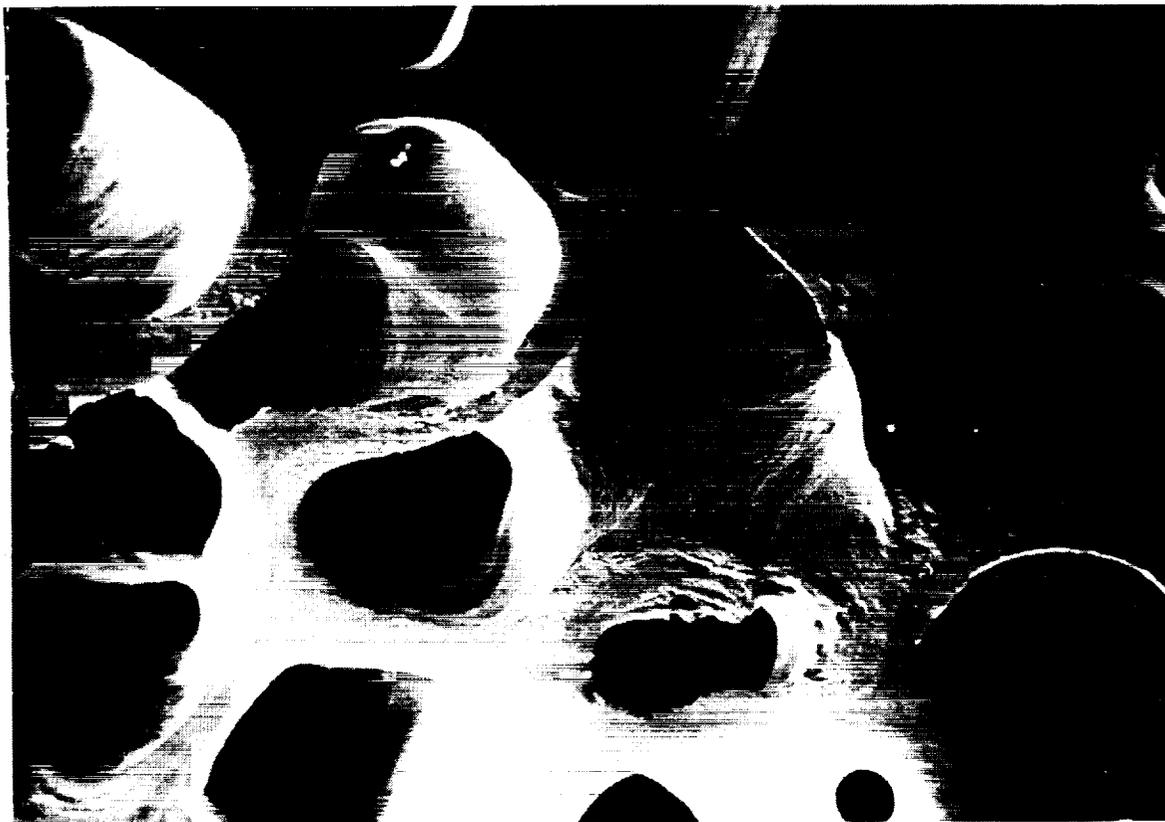


Figure 7a. *Low power scanning electron micrograph of an iliac crest biopsy from a 44-year-old normal male (reproduced with permission).*



Figure 7b. *Low power scanning electron micrograph of a iliac crest biopsy from a 47-year-old osteoporotic female with multiple vertebral compression fractures (reproduced with permission).*

It is known from prolonged bed-rest studies that once the subjects are restored to their normal activity they are capable of restoring their bone mineral density in the os calcis to prebed-rest levels [4]. In Skylab studies follow-up bone density measurements at 3 months showed that the deficit had been repaired in only one astronaut, 50 percent recovery in one, and no recovery in the third who had measured calcaneal loss during space flight [13]. A 5-year follow-up of the mineral content in the os calcis of nine crew members from the Skylab program when compared to eight of their alternates who served as controls also showed a statistically significant loss compared to the control group [18]. The authors cautioned, however, that there are so many potential causes for the observed differences that more studies needed to be done. It is not known whether this restoration would also occur in astronauts and cosmonauts who have experienced microgravity for extended periods such as a 3-year mission to Mars. Furthermore, even if the bone mineral content is restored we do not know whether the three dimensional structure of the bone could be restored to its original condition.

Can Pharmacologic Intervention Regulate Bone Remodeling?

Can you maintain and/or restore bone mineral density while in microgravity? Schneider and MacDonald [14] evaluated a variety of therapies including lower body negative pressure, exercise, static compression, calcitonin, disphosphonate, phosphate, and calcium plus phosphate, and concluded that they were unable to prevent negative calcium balance during bed rest (Figures 8 and 9).

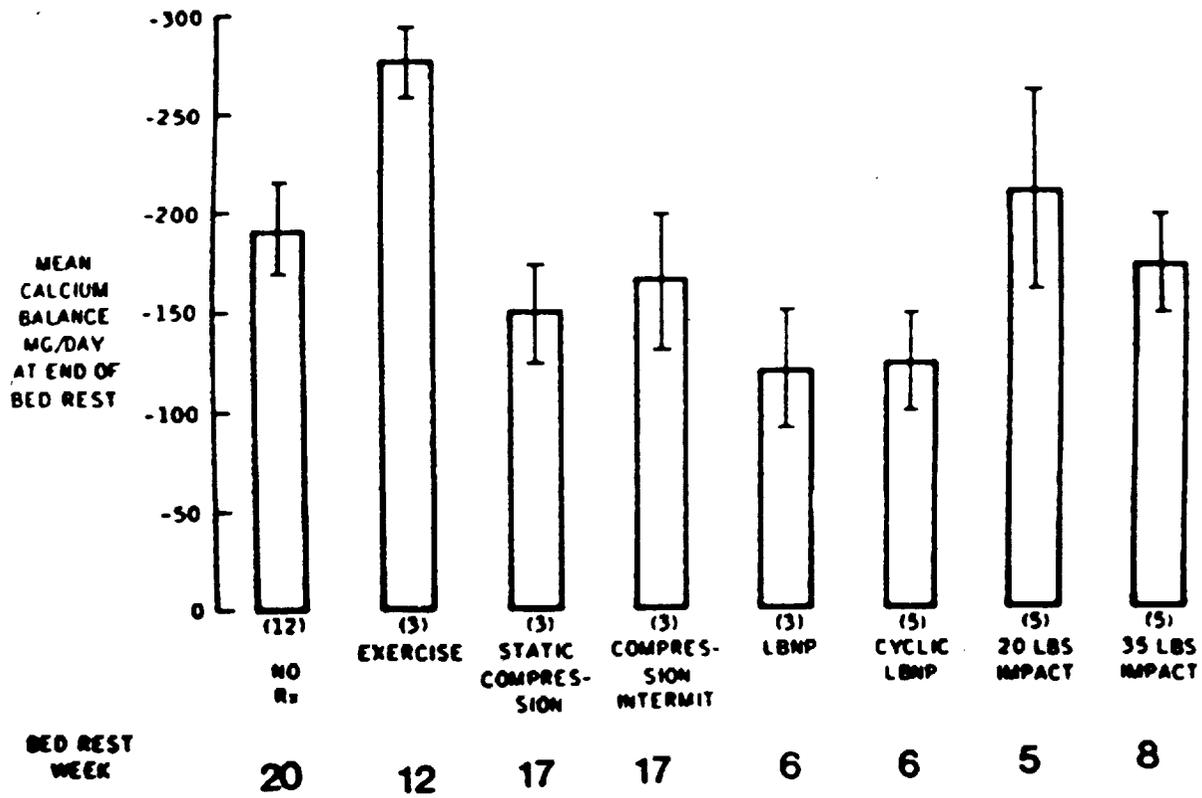


Figure 8. Mean calcium balance of the final week of study during untreated bed rest and in eight physical interventions tried during bed rest (lower body negative pressure [LBNP]) (reproduced with permission).

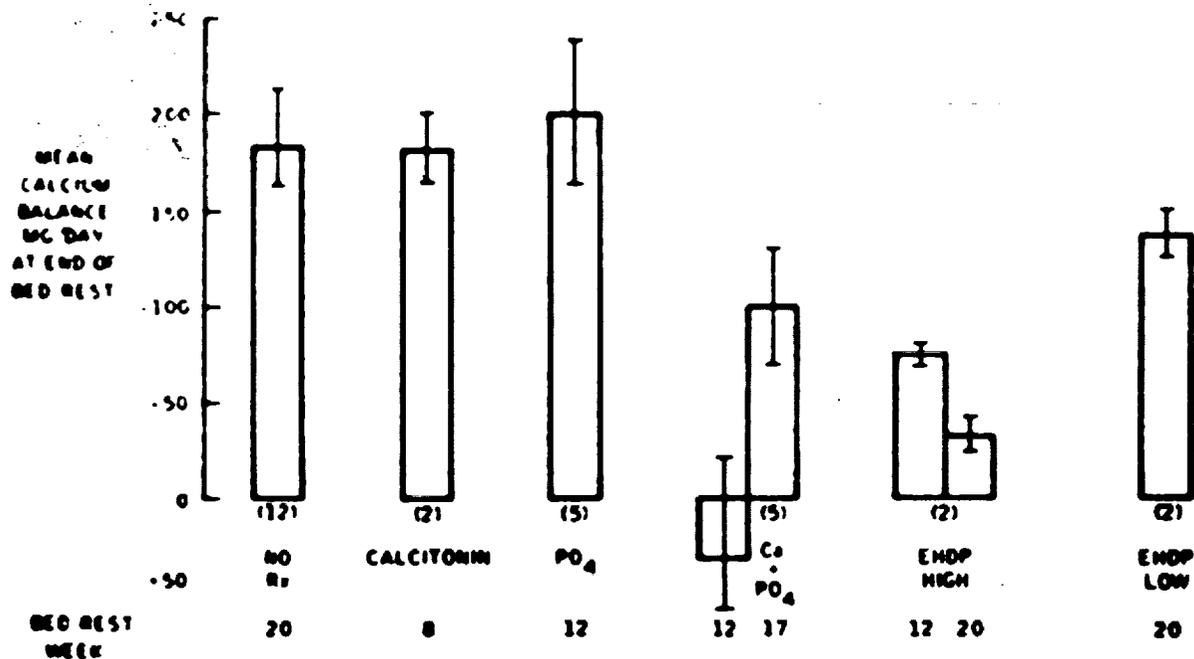


Figure 9. Mean calcium balance during the final week of study during untreated bed rest and in four biochemical therapies tried during bed rest (etidronate [EHDP]) (reproduced with permission).

There is mounting evidence that both $1,25(\text{OH})_2\text{D}$ and parathyroid hormone not only enhance bone calcium mobilization but also act as anabolic hormones to maintain bone remodeling activities. Mature osteoclasts do not possess receptors for either $1,25(\text{OH})_2\text{D}$ or PTH. Instead it is now believed that both of these hormones act on osteoblasts to induce cytokines that regulate osteoclastic activity [6]. Additionally $1,25(\text{OH})_2\text{D}$ and PTH may mobilize stem cells to become mature osteoclasts. Other factors that may play a role in the bone remodeling process include TGF-beta and other bone growth factors. In some studies calcitonin has been shown to increase bone mineral density, but in general it is believed to be an agent to inhibit the bone mobilization and has little effect on the bone remodeling process. Of great interest is the possibility of activating then depressing the bone remodeling activity. This has led to a new cyclic therapy called ADFR (activation, depression, free period, repeat). Although preliminary results suggests that PTH followed by $1,25(\text{OH})_2\text{D}_3$ or phosphate followed by $1,25(\text{OH})_2\text{D}_3$ may significantly increase spinal bone density [15], Schneider and MacDonald [14] have reported that during bed rest calcitonin, phosphate, or diphosphonate therapy did not significantly reverse the negative calcium balance although some improvement was found in the group receiving high amounts of diphosphonate. Recently Judge *et al.* [7] evaluated bone biopsies from four male volunteers after 17 weeks of bed rest and found a mismatch between bone resorption and bone formation, whereby bone resorption continued or was increased while bone formation was decreased. Arnaud *et al.* [2] evaluated bone biopsies from volunteers after a 2-week period of bed rest. After double tetracycline labeling, it was found that bone formation rates significantly diminished in six

subjects, was unchanged in one, and higher in one subject who was a swimmer (personal communication).

Can Bone Cells Adapt to Microgravity?

Little is known about the impact of microgravity on bone cell activity. Can bone cells adapt to a microgravity environment? Are bone cells addicted to gravity and therefore require gravity to maintain normal bone remodeling activity? An insight into these questions has recently become available as a result of a study on board STS-59. Sixteen fertilized chicken eggs at gestational ages of 2 and 9 days were housed in a incubator and flown on board the Space Shuttle Discovery. Control eggs of the same gestational ages were maintained in a similar incubator and exposed to the vibrations and jostling to simulate liftoff and reentry. After the 5-day flight one-half of the eggs from both groups were immediately sacrificed. The 2-day old embryos that were flight flown all stopped developing and died within 24 hours, whereas all of the 2-day old control embryos survived. The flight flown 9-day old embryos all developed similar to the control group. One-half of the flight flown and control embryos had their bones evaluated. Preliminary results suggest that the bone modeling process in the flight flown animals was no different from the earth controls [19]. These results suggest that microgravity does not alter the bone mineralization process. Therefore, it is likely that bone cells become adapted to gravity. Once exposed to microgravity they decrease their activity. Thus, if we could understand what the signal or signals are that microgravity provides to bone cells to limit their activity, it may be possible to reverse this action. Alternatively it may be that if humans are born in a microgravity environment that their bone cells would develop normally and a normal skeleton would evolve. As we contemplate exploring new solar systems it may be that there will be a need for microgravity born astronauts to complete such missions.

Conclusion

The likely physiologic events in calcium and bone metabolism that occur in a microgravity environment are as follows:

1. The unloading of the G-forces on the skeleton induce or inhibit a signal(s) that causes a depression in bone formation and an acceleration in bone demineralization that results in an increase in circulating ionized calcium concentrations.
2. The small rise in ionized calcium concentrations is recognized by the parathyroid glands, which in turn decrease their secretion of parathyroid hormone.
3. The decrease in parathyroid secretion causes an increase renal loss of calcium into the urine.
4. A decrease in PTH secretion also results in the depression of the metabolism of 25-hydroxyvitamin D (25-OH-D) to 1,25-dihydroxyvitamin D (1,25[OH]₂D) [5, 6].

The decrease in production of 1,25(OH)₂D results in a decrease in the efficiency of intestinal calcium transport (Figure 10). This cycle ultimately results in the continued wasting of the bone mineral.

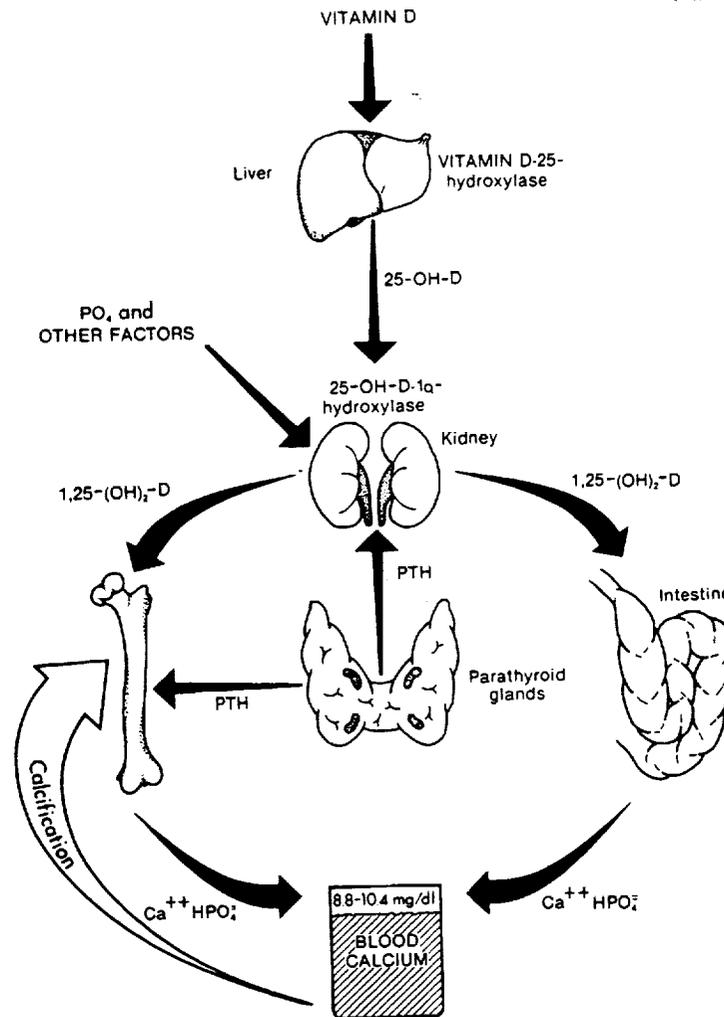


Figure 10. *Schematic representation of the hormonal control loop for vitamin D metabolism and function. A reduction in the serum calcium below approximately 8.8 mg/mL prompts a proportional increase in the secretion of parathyroid hormone, which enhances the mobilization of calcium stores from bone. Parathyroid hormone also promotes the synthesis of 1,25(OH)₂D in the kidney, which in turn stimulates the mobilization of calcium from bone and its absorption from the intestine (reproduced with permission).*

We do not know whether microgravity in enhancing the mobilization of calcium from bone also causes the loss of bone matrix. If the matrix is maintained there is a good possibility that astronauts can stimulate bone mineralization in a microgravity environment during prolonged space travel. If, however, gravity adapted bone cells are further stimulated during astronaut training to become more dependent on gravitational forces then the acute loss of Earth's G-force could accelerate the mobilization of calcium from the bones of astronauts. A detraining period of the skeleton may be an alternative approach to limiting the total amount of calcium lost from bone during exposure to microgravity.

References

- [1] ARNAUD, S. B, V. S. SCHNEIDER, AND E. MOREY-HOLTON. Effects of inactivity on bone and calcium metabolism. In: *Inactivity: Physiological Effects*, edited by H. Sandler and J. Vernikos. London: Academic Press, Inc., 1986, p. 49-76.
- [2] ARNAUD, S. B., D. J. SHERRARD, N. MALONEY, R. T. WHALEN, AND P. FUNG. Reduced bone formation is measurable in the iliac crest of normal men after 7 days in a bed rest model of weightlessness. *J. Bone Min. Res.* 4:s233, 1989. (Abstract)
- [3] DEMPSTER, D. W., E. SHANE, W. HORBERT, AND R. LINDSAY. A simple method for correlative light and scanning electron microscopy of human iliac crest bone biopsies: qualitative observations in normal and osteoporotic subjects. *J. Bone Min. Res.* 1:15-21, 1986.
- [4] DONALDSON, C. L, S. B. HULLEY, J. M. VOGEL, R. S. HATTNER, J. H. BAYERS, AND D. E. MCMILLIAN. Effect of prolonged bed rest on bone mineral. *Metabolism* 19:1071-1084, 1970.
- [5] HOLICK, M. F. Vitamin D and the kidney. *Kid. Internatl.* 32:912-929, 1987.
- [6] HOLICK, M. F. Vitamin D: biosynthesis, metabolism, and mode of action. In: *Endocrinology, Volume 2*, edited by L. J. DeGroot. New York: Grune & Stratton, 1989, p. 902-926.
- [7] JUDGE, D., V. S. SCHNEIDER, AND A. LEBLANC. The histomorphometric changes in disuse osteoporosis. *J. Bone Min. Res.* 4:S238, 1989. (Abstract)
- [8] KRANE, S. M. AND M. F. HOLICK. Metabolic bone disease. In: *Harrison's Principles of Internal Medicine, 11th Edition*, edited by E. Braunwald, K. H. Isslebacher, R. G. Petersdorf, J. S. Wilson, J. B. Martin, and A. S. Fauci. New York: McGraw-Hill, 1986, p. 1889-1900.
- [9] LEBLANC, A., V. S. SCHNEIDER, J. KREBS, H. EVANS, S. JHINGRAN, AND P. JOHNSON. Spinal bone mineral after 5 weeks of bed rest. *Calcif. Tissue Int.* 41:259-261, 1987.
- [10] PARFITT, A. M. Bone effects of space flight: analysis by quantum concept of bone remodeling. *Acta Astronautica* 8:1083-1090, 1981.
- [11] RAMBAUT, P. C., C. S. LEACH, G. D. WHEDON. A study of metabolic balance in crewmembers of Skylab 4. *Acta Astronautica* 6:1313-1322, 1979.
- [12] RAMBAUT, P. C. AND R. S. JOHNSTON. Prolonged weightlessness and calcium loss in man. *Acta Astronautica* 6:1113-1122, 1979.
- [13] SCHNEIDER, V. S., A. LEBLANC, AND P. RAMBAUT. Bone and mineral metabolism. In: *Space Physiology and Medicine*, edited by A. Nicogossian, C. Huntoon, and S. Pool. Philadelphia: Lee & Febiger, 1989, p. 214-221.
- [14] SCHNEIDER, V. S. AND J. MCDONALD. Skeletal calcium homeostasis and countermeasures to prevent disuse osteoporosis. *Calcif. Tissue Int.* 36:S151-S154, 1984.
- [15] SLOVIK, D. M., D. I. ROSENTHAL, S. H. DOPPELT, J. T. POTTS, M. A. DALY, J. A. CAMPBELL, AND R. M. NEER. Restoration of spinal bone in

- human parathyroid hormone (1-34 and 1,25-dihydroxyvitamin D.
Res. 1:377-381, 1986.
- [16] STEWARD, A. F., M. ADLER, C. M. BYERS, G. V. SEGRE, AND A. E. BROADUS. *N. Engl. J. Med.* 306:1136-1140, 1982.
- [17] STUPAKOV, G. P., V. S. KAZEYKIN, A. P. KOZLOVSKIY, AND W. KOROLEY. Evaluation of changes in human axial skeletal bone structures during long-term space flights. *Kosm. Biol. Med.* 18:33-37, 1984.
- [18] TILTON, F. E., J. J. C. DEGIOANNI, AND V. S. SCHNEIDER. Long-term follow-up of Skylab bone demineralization. *Aviat. Space, Environ. Med.* 51:1209-1213, 1980.
- [19] VELLINGER, J., M. DEUSER, M. CASTONGUAY, H. KAVOOKJIAN, D. FERMIN, R. KERSCHMANN, S. HESTER, R. HULLINGER, S. HUNTER, AND M. F. HOLICK. Effect of weightlessness on bone development and mineralization in chick embryos on board the Space Shuttle Discovery. *J. Bone Min. Res.* 4:s409, 1989. (Abstract)
- [20] VICO, L., D. CHAPPARD, C. ALEXANDRE, S. PALLE, P. MINAIRE, G. RIFFAT, B. MORUKOV, S. RAKHMANOV, AND V. S. OGANOV. Quantitative histological study of the bone mass and cellular activities after a 120-day period of decubitus. Test of preventive protocols. *Ann. Biol. Clin. (Paris)* 45:145-151, 1987.